

An Investigation of the Palatability of Some Marine Invertebrates to Four Species of Fish

ELEANOR RUSSELL¹

IN THE PAST, fish attractants have aroused attention, and "bait" extracts have been made which attract fish and excite them to feeding activity (Bateson, 1890; van Weel, 1952; and Steven, 1959).

Less work has been done on repellent substances, apart from shark repellents such as copper acetate (Whitley and Payne, 1947). The repelling action of copper acetate may be due to acetic acid, and it has long been known that dilute acids are repellent to fish (Bateson, 1890, confirmed by Hiatt, Naughton, and Matthews, 1953).

Fish accept eagerly some animal baits while completely ignoring others. On this basis, different degrees of "palatability" ranging to "unpalatability" might be postulated. Among known unpalatable animals there are no obvious common factors with regard to colour or shape, and this suggests that unpalatability is based on the chemical senses.

Many soft-bodied invertebrates are openly exposed to fish attack, and even in the absence of obvious protective devices remain unmolested. Some workers (Garstang, 1890*a, b*; Bullock, 1955; Thompson and Slinn, 1959; Thompson, 1960*a*) have shown that certain marine animals make themselves distasteful to predators by the secretion of diffusible "repellent" substances.

The present investigation was undertaken to obtain data on the relative attractiveness to fish of a diversity of marine animals in southern Queensland. Comparisons of food preferences were made, following in principle the methods of earlier work by Stephenson and Grant (1957) and Blaxter and Holliday (1958).

¹ Present Address: Zoology Department, University of New South Wales, Sydney, Australia. Manuscript received May 25, 1965.

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GENERAL METHODS

Test Animals

Two marine species, *Pelates quadrilineatus* (Bloch), trumpeter, and *Torquigener hamiltoni* (Gray and Richardson), toado; and two freshwater species, *Gambusia affinis* Baird and Girard, and *Carassius auratus* (L.), goldfish, were used. *Carassius* was obtained from a dealer and the other three species were netted when required.

Freshwater species were used as a control against the familiarity of marine species with invertebrates of their own environment.

Aquarium Conditions

Two glass aquaria (61 × 30.5 × 30.5 cm) without sand were used for each species. Weed was excluded because fish may eat it. Approximately 20 mussels, *Trichomya hirsutes* (Lamarck), were kept in each marine aquarium to remove sediment. Aeration was continuous and salinities were checked weekly. Sizes of the fish, their dietary types, and numbers in aquaria are given in Table 1.

Maintenance Feeding

Fish began eating after a few days and were maintained on prawn, *Metapenaeus mastersii* (Haswell). Each day food was dropped on the

water. Particles were seized before they reached the bottom.

A known amount of food was introduced, and the amount remaining uneaten after 2 hr was removed and weighed after excess water was removed. This gave a rough measure of the amount of food to be fed each day. Table 1 gives food consumption in gm/gm fish/week.

Testing Methods

Feeding trials were made each day before normal feeding. During tests less normal food (prawn) was given to allow for other food eaten. Only two or three unpalatable materials could be tested in one series of tests, i.e. within 1 hr. If more were tested, the fish ignored even normally palatable material for 3–4 hr.

Comparisons of palatability were made by dropping simultaneously two equal-sized fragments on the water's surface as distant from the fish as possible. As soon as the particles hit the water, the fish swam towards them. Usually both particles were falling through the water by the time the fish reached them. The reaction of the fish to each fragment was observed, and an estimate made of which was preferred. The size of pieces used was that most suitable for the species (see Experiment I). Tests were repeated twice where possible, with an interval of at least one day between tests.

Anticipatory responses were soon evident

whenever a person approached an aquarium. This was particularly so with *Gambusia* and *Carassius*. It was considered that this response would not affect the results: while the initial attraction was visual, final acceptance depended on other factors.

EXPERIMENT I

GENERAL REACTIONS TO INTRODUCED MATERIAL

The reactions of each species to one palatable material (*Metapenaeus*), and to one "neutral," inedible material (plastic tubing) was tested. The significance of different particle size for later tests of palatability was noted.

Results

Gambusia reacted to a large piece of *Metapenaeus* by clustering densely round it and biting at it vigorously for up to 30 minutes. Fragments less than 4 mm across were eaten too rapidly by one or two fish to allow proper classification of palatability.

Gambusia investigated briefly anything dropped into the water. They swam up to a piece of plastic tubing, bit at it a few times, and then let it fall to the bottom where it was ignored.

Carassius swallowed immediately small fragments (less than 4 mm across) of any material.

TABLE 1
DETAILS OF TEST ANIMALS EMPLOYED

SPECIES	SIZE OF FISH (cm)	DIETARY TYPE	AQUARIUM DENSITY: NO. OF FISH PER TANK	AMOUNT OF FOOD NORMALLY GIVEN/WEEK
<i>Gambusia</i>	2.5–3.5	omnivorous	20–30	0.03 gm/gm
<i>Carassius</i>	5–6	omnivorous (Suyehiro, 1942)	4–6	0.035 gm/gm
<i>Pelates</i>	6–9	no published data; omnivorous in aquaria.	4–6	0.2 gm/gm
<i>Torquigener</i>	6–9	no published record; omnivorous in aquaria. Suyehiro (1942) records two related spp. as omnivorous.	4–6	0.2 gm/gm

Small pieces of material such as plastic tubing were rejected some time later. The time of retention was so variable that large fragments were always given for palatability tests. *Carassius* continued to suck at a large piece of *Metapenaeus* for up to 30 minutes. They sucked at a large piece of plastic tubing for up to 20 seconds and then ignored it.

Pelates and *Torquigener* rapidly tore up large pieces of *Metapenaeus*. They bit at large pieces of plastic tubing once or twice and then ignored them. Small pieces were sometimes swallowed and ejected later, so large pieces were always used for tests of palatability.

Any material allowed to remain on the floor of the tank was investigated from time to time by all fish.

EXPERIMENT II

COMPARISONS OF PALATABILITY

Fragments of the tissues of a number of marine invertebrates were fed directly to fish in the comparison situation described above. Tissue of unknown palatability was compared with a known tissue. The initial standard was *Metapenaeus*, but when this proved to be more highly palatable than any other tissue tried, a less palatable substandard was adopted, *Pyrazus ebeninus* Brugière (whelk). The palatability of tissue from 48 different species was tested in this way. Because of the subjectivity of the method, three levels of palatability have been defined on the basis of the reactions of each fish to different types of material. Each material has been assigned to a particular level, rather than placed on an absolute scale.

Results

Three levels of palatability have been defined on the basis of palatability comparisons:

(1) Highly palatable (++) material was always eaten very rapidly by *Pelates* and *Torquigener*, with no preliminary sampling. *Gambusia* and *Carassius* clustered about the material and continued to tear pieces from it for more than 10 minutes and up to 30 minutes.

(2) Palatable (+) material was sampled for a short time before being eaten by *Pelates* and *Torquigener*. *Gambusia* and *Carassius* clustered about the material and tore pieces from it for

a much shorter time, less than 10 minutes, and generally less than 5 minutes.

(3) Unpalatable (0) material was not eaten, but the fish investigated it and often mouthed at it. Differences in degree of unpalatability were noticed: some material was swallowed and later ejected, some material was never swallowed.

Table 2 lists the palatability of all materials tested in this experiment. Minor differences between the four fish in the position of test materials on a nominal scale were not considered significant. There were no differences in the assignment of materials tested to any level of palatability.

Table 3 shows all materials, classed as palatable and unpalatable, grouped with respect to colour. A chi-squared test (Siegel, 1956) shows that the differences in colour distribution between palatable and unpalatable are significant ($\chi^2 = 16.57$, $P < .001$).

EXPERIMENT III

COMPARISONS OF PALATABILITY WHEN TEXTURE AND CHARACTERISTIC APPEARANCE ARE DISGUISED

In Experiment II, no examples of unpalatability were found where appearance was obviously responsible (since all materials were "tasted" before being rejected), but texture was believed to be of some importance. For these tests 13 unpalatable animals were selected to determine palatability after texture was disguised and general appearance was altered.

A solution of gelatin in sea water was used as a base material to which test materials were added. A final concentration of 20% gelatin produced a gel stiff enough to be cut into small pieces which dissolved only slowly in water. The reaction of the fish to gelatin was the same as their reaction to plastic tubing. A homogenate of *Metapenaeus* was added in concentrations of 10–50% to render the whole palatable. A concentration of 10% (by weight) of *Metapenaeus* was sufficient to make the gelatin block highly palatable, and the reaction to 10% *Metapenaeus* was indistinguishable from the reaction to 50% *Metapenaeus*.

Tissues for testing were stored in a deep freeze after collection, and ground in a blender.

TABLE 2
RELATIVE PALATABILITY TO FISH OF ORGANISMS TESTED

MATERIAL TESTED	NO. OF TRIALS	PART OF ANIMAL USED	<i>Gambusia</i>	<i>Carassius</i>	<i>Pelates</i>	<i>Torquigener</i>
CRUSTACEA						
<i>Metapenaeus mastersii</i> (Haswell)	721	tail muscle	++	++	++	++
<i>Callinassa australiensis</i> Dana	1	tail muscle	++	++	++	++
<i>Mictyris longicarpus</i> Latreille	2	leg muscle	++	++	++	++
<i>Alpheus</i> sp.	2	tail muscle	+	+	+	+
ANNELIDA						
<i>Marphysa sanguinea</i> (Montagu)	4	body segments	++	++	++	++
<i>Onuphis teres</i> (Ehlers)	2	body segments	++	++	++	++
<i>Phyllodoce malgremi</i> Cuvier	2	body segments	++	++	++	++
<i>Pheretima</i> sp.	3	body segments	++	++	++	++
<i>Thelepus setosus</i> (Quatrefages)	3	body segments	+	+	+	+
<i>Thelepus setosus</i> (Quatrefages)	2	tentacles	+	+	+	+
<i>Clymene grossa</i> var. <i>tropica</i> Monro	2	body segments	+	+ → 0	+ → 0	+ → 0
Unidentified terebellid	1	body segments	+	+ → 0	+ → 0	+ → 0
<i>Audouinia tentaculata</i> (Montagu)	3	body segments	0	0	0	0
ECHIURIDA						
<i>Ochetostoma australis</i> Edmonds	2	body wall	+	+	+	+
SIPUNCULOIDEA						
<i>Phascolosoma dunwichi</i> Edmonds	2	body wall	+	+	+	+
MOLLUSCA						
Lamellibranchiata						
<i>Tricbomya hirsutes</i> (Lamarck)	5	mantle	+	+	+	+
<i>Crassostrea commercialis</i> (Iredale & Roughly)	4	mantle	+	+	+	+
Cephalopoda						
Squid	2	muscle	+	+	+	+
Gastropoda						
<i>Uber plumbeum</i> Lamarck	3	foot	+	+	+	+
<i>Pyrazus ebeninus</i> Brugiere	10	foot	+	+	+	+
<i>Nassarius luridus</i> (Gould)	4	foot	+	+	+	+

The organisms tested are arranged according to animal group. Within groups they are arranged in approximate order of palatability.

TABLE 2 (Continued)

MATERIAL TESTED	NO. OF TRIALS	PART OF ANIMAL USED	<i>Gambusia</i>	<i>Carassius</i>	<i>Pelates</i>	<i>Torquigener</i>
<i>Nerita chameleon</i> Linn.	2	foot	+	+	+	+
<i>Austrococblea obtusa</i> (Dillwyn)	1	foot	+	+	+	+
Opisthobranchiata						
<i>Phyllidea pustulosa</i> Cuvier	2	foot	0	0	0	0
<i>Onchidina australis</i> Semper	2	foot	0	0	0	0
<i>Glossodoris festiva</i> (Adams)	4	foot	0	0	0	0
<i>Aplysia angasi</i> (Sowerby)	3	foot	0	0	0	0
<i>Dolabella</i> sp.	13	foot	0	0	0	0
<i>Notarchus leachii</i> Blainville	4	foot	0	0	0	0
<i>Umbraculum sinicum</i> Gmelin	2	foot	0	0	0	0
BRACHIOPODA						
<i>Lingula bancroftii</i>	2	"stem"	+	+	+	+
Johnston and Hirshfeld	2	body	+	+	+	+
BRYOZOA						
Red colonial	2	part of whole body	0	0	0	0
ECHINODERMATA						
Asteroidea						
<i>Antbenea</i> sp.	3	arms and tub. feet	+	+	+	+
	2	hepato-pancreas and gonads	+	+	+	+
HOLOTHUROIDEA						
Holothuroidea						
<i>Holothuria scabra</i> Jager	2	body wall	0	0	0	0
<i>Holothuria leucospilota</i> (Brandt)	3	body wall	0	0	0	0
<i>Holothuria pardalis</i> Selenka	2	body wall	0	0	0	0
NEMERTEA						
Black, with yellow stripe	2	part of whole body	0	0	0	0
PORIFERA						
Yellow	1	part of whole body	0	0	0	0
Black	2	part of whole body	0	0	0	0
COELENTERATA						
<i>Alcyonium</i> sp.	5	part of whole body	0	0	0	0
Blue soft coral (fam. Xenidae)	2	part of whole body	0	0	0	0
Sea Pen	1	part of whole body	0	0	0	0

TABLE 2 (Continued)

MATERIAL TESTED	NO. OF TRIALS	PART OF ANIMAL USED	<i>Gambusia</i>	<i>Carassius</i>	<i>Pelates</i>	<i>Torquigener</i>
<i>Catostylus mosaicus</i>	2	homogenate of whole	0	0	0	0
<i>Cerianthus</i> sp.	3	base	0	0	0	0
	3	tentacles of live animal	0	0	0	0
	2	tentacles of dead animal	0	0	0	0
Other anemone	1	tentacles of dead animal	0	0	0	0
ENTEROPNEUSTA						
<i>Glossobalanus</i> sp.	4	anterior part of body	0	0	0	0
PISCES						
<i>Sillago maculata</i>						
Quoy & Gaimard	2	muscle	0	0	0	0
<i>Torquigener hamiltoni</i> (Gray & Richardson)	3	muscle	0	0	0	0

In the preparation of gelatin-base mixtures, a mixed homogenate of *Metapenaeus* and of test tissue was combined with an equal weight of 40% gelatin in sea water to give a final concentration of 20% gelatin. *Metapenaeus* + test tissue mixtures were made in the following proportions:

METAPENAEUS	TEST TISSUE	FINAL CONC. OF TEST TISSUE (%)
5 parts	0 parts	0
4 parts	1 part	10
3 parts	2 parts	20
2 parts	3 parts	30
1 part	4 parts	40
0 parts	5 parts	50

(A criticism of this method is that the concentration of *Metapenaeus* alters as well as that of the test tissue. But if the *Metapenaeus* concentration is kept constant, the total concentration of tissue, and therefore of texture, is altered.)

Within each series, tests were made to find the concentration of test tissue required to make the whole unpalatable. This was checked by comparison with the concentrations above and

below. Tests of different parts or secretions of some animals were also made in this way.

Results

Table 4 lists the animals tested and the concentration necessary to make the mixture of prawn and gelatin unpalatable. Materials are arranged in three groups; the difference between the two "slightly unpalatable" animals and the others is probably real, but the significance of the difference in effect between concentrations of 20% and 30% of test tissue could be questioned.

In no case was there any noticeable difference in palatability between different parts of an animal. When the fresh purple ink of the opisthobranchs *Notarchus* and *Dolabella* was introduced into the fish tanks, the fish swam towards it and through it, often biting at local higher concentrations.

EXPERIMENT IV

SURFACE pH

Thompson and Slinn (1959) and Thompson (1960*a, b*) have shown that some molluscs produce a defensive acid external secretion

which renders them unattractive to fish. Since the five molluscs tested in Experiment III were all highly unpalatable, their surface pH when alive was measured, and that of all the other animals which were tested in Experiment III.

Universal pH paper was used to measure the normal surface pH after the animal had been placed in a glass dish and allowed to drain for five minutes. Then the surface was wiped with a filter paper, stimulated gently with a glass rod, and the pH ions remeasured.

Results

The surface pH in all cases was approximately 7.0. This result by no means rules out the possibility of acid secretion, but continuous acid secretion and secretion after gentle disturbance are unlikely.

DISCUSSION

Of 48 species tested, the 24 which were unpalatable to fish came from a wide range of animal groups.

It is difficult to compare these results in detail with those of other workers. Stephenson and Grant (1957) worked on *Plectroplites ambiguus* (Richardson), and marine molluscs occupied an equivalent position in the preference scale. However, marine Crustacea were less attractive for *Plectroplites* than were some

marine molluscs, and the most attractive foods for *Plectroplites* (freshwater crustaceans and squid) were relatively less palatable in the present investigation.

Steven (1959) compared the relative effectiveness of natural food substances in evoking exploratory feeding behaviour in *Hepsitia stipes* (Muller and Tröschel) and in *Bathystoma rimator* (Jordan and Swain). Extracts of fresh plankton and of the muscle of the bivalve *Arca zebra* were effective, while the echinoids *Diadema antillarum* and *Lytechnis variegatus* and a holothurian, *Holothuria* sp., also produced intense responses from less extreme dilutions. In the present work certain species of *Holothuria* were found to be unpalatable to fish. However, the presentation of a very small amount of food material, whether palatable or unpalatable (e.g., *Holothuria*), caused exploratory feeding reactions which lasted for one to two minutes.

Three main factors could influence unpalatability: (1) "taste" due to diffusible chemical substances; (2) texture (though not unattractive, the organism may be hard or tough, or may contain spicules); (3) colour, either in itself, or due to association with some other unpleasantness.

In Experiment III, it was shown that unpalatable animals remained unpalatable when the texture was completely altered, and colour to some extent was altered. If colour was the sole cause of unpalatability, fish would ignore food of some particular colour without going near it or taking it into their mouths. Results indicate that this is not generally so; and, while Table 3 indicates that differences in colour between palatable and unpalatable animals are significant, there must be other factors contributing to unpalatability.

Many sedentary, soft-bodied, "unprotected" animals are unpalatable, and these include Porifera, Coelenterata, Nemertea, some Polychaeta, Opisthobranchiata, and Holothuroidea. Many of these animals have no obvious protective devices: *Holothuria scabra* is just as unpalatable as *H. leucospilota*, but has no Cuvierian tubules; a few opisthobranchs eject purple ink, but many do not. Porifera and Nemertea have no obvious physical means of

TABLE 3

EFFECT OF COLOUR ON PALATABILITY:
RELATION BETWEEN COLOUR AND DEGREE OF
PALATABILITY

COLOUR	HIGHLY PALATABLE AND PALATABLE (NUMBER)	UNPALATABLE (NUMBER)
White)		
Grey)	14	2
Cream)		
Red)		
Yellow)	8	13
Orange)		
Brown)		
Blue)		
Green)	3	5
Purple)		
Black	0	3

TABLE 4
TESTS OF PALATABILITY WHEN TEXTURE AND COLOUR ARE MASKED BY GELATIN

GROUP	TEST SPECIES	PART OF BODY TESTED	CONCENTRATION NECESSARY TO MAKE GELATIN PLUS PRAWN UNPALATABLE
1 (Slightly unpalatable)	<i>Clymene</i>	whole animal	50%
	<i>Cirratulus</i>	whole animal	50%
2 (Unpalatable)	<i>Torquigener</i>	muscle	30%
	<i>Holothuria</i>	whole animal	30%
	<i>pardalis</i>	cuvierian tubule	30%
	Black sponge	whole animal	30%
		mucus from surface	30%
	<i>Catostylus</i>	whole animal	30%
	<i>Alcyonium</i> sp.	whole animal	30%
3 (Highly unpalatable)	Nemertine	whole animal	20%
	<i>Glossodoris</i>	whole animal	20%
	<i>Aplysia</i>	mantle and foot	20%
	<i>Notarchus</i>	mantle and foot	20%
	<i>Dolabella</i>	mantle and foot	20%
	<i>Umbraculum</i>	mantle and foot	20%

protection, although the texture of a sponge may be sufficient protection.

The most unpalatable animal seemed to be the pleurobranch *Umbraculum*, which most fish did not touch. McNae (1962) notes that "when handled the animal (*Umbraculum*) gives off a characteristic scent reminiscent of that of aplysiids. This scent may have some defensive function." The author has noted a characteristic odour in many opisthobranchs, notably *Aplysia* and *Glossodoris*.

Thompson and Slinn (1959) noted an acid secretion which was apparently responsible for making *Pleurobranchus* distasteful to predators. They recorded that the predators "tasted" but discarded it immediately and, in the case of fish, often violently. Although *Pleurobranchus* is brightly coloured, this did not seem to warn the fish in any way.

In the present experiments, animals with bright, supposedly warning, colouration remained unpalatable when their pattern was destroyed and their colour partly masked by gelatin and *Metapenaeus*. Some cryptically coloured, well camouflaged animals were also unpalatable, notably *Dolabella*, *Notarchus*, *Aplysia*, *Holothuria scabra*, and *H. pardalis*.

These experiments and those of Thompson (1960*a, b*) do not support the idea which seems to have originated with Herdman (1890*a, b*), Garstang (1889, 1890), and Crossland (1911) that cryptically coloured, camouflaged animals were acceptable to fish as food, while brightly coloured ones were rejected because of their warning colouration.

While the results point to the importance of chemical defense mechanisms, the presence of an acid secretion does not seem to be the cause of unpalatability in any of the animals whose external pH was measured. Of all the gastropods tested by Thompson (1960*b*), only five produced an acid secretion, while the remainder had similar glands in the epidermis which produced a secretion which presumably also had a defensive function. In this connection, one must mention the "characteristic odour" of *Umbraculum*, *Aplysia*, and *Dolabella*.

The complex phenomena of learning must play some part in the selection of food by fish, and it is at this level that colour may be important, but learning cannot determine the causes of unpalatability. It is suggested that chemical defense mechanisms may be widespread among invertebrates and that a search

for these, in addition to more obvious mechanisms, would be rewarding.

SUMMARY

1. The palatability of 48 species of marine invertebrates has been determined, of which 24 species were found to be unpalatable to the four test species of fish.

2. There is a significant difference in colour distribution between palatable and unpalatable tissues.

3. Unpalatability remained when texture, colour pattern, and to some extent colour, were masked by presenting the tissue in a gelatin base.

4. It is suggested that chemical defense mechanisms are widespread among invertebrates, and an important factor in determining what fish eat.

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